

Brazilian mycotoxin experiences

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Throughout the last 30 years, Brazilian swine production chains have experienced great evolution due to research and development in fields such as nutrition, health, genetics, and management. However, there are still some hindrances for those chains.

Brazil also became one of the largest producers and exporters of grains, given the vast tracts of farmland and the significant investments in technologies, which culminated with the record production of grains in 2010 estimated in 149.5 million tons, especially in planting corn and soybeans.

In the area of feed production, the volume of grain produced represented approximately 60 million tons of ration, working with approximately US\$ 22 billion.

However, due to the predominance of tropical and subtropical climate, characterised by temperature oscillations and by high humidity, coupled with the difficulty of drying and storage grains, fungal growth and mycotoxin proliferation occurs with relative ease in cereals, both during the period that they are still in the field, and during the storage or processing.

Considering this fact, it is important to implement programs for fungi and mycotoxin contamination control in grains and foods. Among the 140 thousand matrices analysed at LAMIC in 25 years for the detection of mycotoxins, about 55% were contaminated with at least one type of mycotoxin.

Moreover, as an example, the presence of aflatoxins in samples of different matrices, mainly maize and derivatives, have been evaluated and over 36% of these samples were positive, with a contamination average of 8.7mg/kg.

Strategies for control

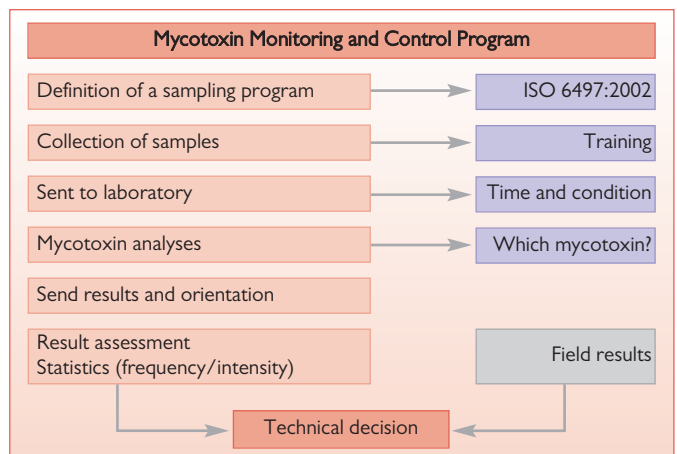
In order to be effective, the strategy for mycotoxicoses control management (Fig. 1) must have an adequate monitoring system.

In Brazil, considering the size of the country and territorial limitations on access to transport services fast enough, access to official laboratories is hindered.

For this reason, there has been increasing use of fast tests, due to the speed for obtaining information.

The improvement of technical training and information availability, allows the diagnosis and taking technical decisions on the efficient routine control of production, especially pigs. It is therefore feasible, for example, by deciding whether or

Fig. 1. Flowchart from the mycotoxin monitoring plan.



Mycotoxin	Products evaluated	Approved	%
Fumonisin	13	7	54
Aflatoxins	12	4	33
Zearalenone	53	11	21

Table 1. Results of anti-mycotoxin additives (AMAs) evaluated from swine 2005-2011.

not anti-mycotoxin additives should be used.

In function of the large production of grains and considering the ease of contamination by fungi producing mycotoxins, some tools for prevention of fungal growth and of mycotoxin contamination have been developed, such as continuous monitoring of grains and foods produced in feed mills and more investment in genetics to develop a more resistant plant to the development of fungi.

However, with regard to genetic improvement of grain, results are still not significant.

In order to optimise the control of raw materials and food in the presence of mycotoxins, LAMIC established logistics of receiving samples and fast analysis, with ready availability of analytical results, so that the livestock industry can manage with security the mycotoxicological problems.

The improvement of the workforce, analytical equipment, and test techniques allowed certification and accreditation of LAMIC by official organisations to conduct several mycotoxins tests.

Among measures that can be adopted to decrease toxic effects of mycotoxins in animal, especially pigs, it is important to highlight the addition of anti-mycotoxin additives (AMA) to the diet of these animals.

The AMA were initially called adsorbents, due to the attraction and adsorption that occurs between the aluminosilicate (one of the first AMA) and to Aflatoxin B1.

The strategy of adsorbing/inactivate mycotoxins in passage through the digestive tract today is the most feasible alternative, being used worldwide in order to decrease the toxic effect of mycotoxins.

Considering the high prevalence of mycotoxins in produced food and characteristics of the agribusiness beyond high intake of AAM, it is essential a careful evaluation of the quality of available products, thus enabling the choice of products really effective in minimising the effects of mycotoxicoses in animals.

Today in Brazil, there is a wide range of products called AMA, which do not have any certificate of evaluation, both in vitro and in vivo.

However, the Brazilian Department of Agriculture have settled some requirements to register and approve this kind of additive and require products to be tested under

specific in vitro and in vivo conditions.

Anti-mycotoxin additives

AMA are inert substances that either bind to mycotoxins, making them unavailable to intestinal absorption, or biotransform them, changing their molecules into atoxic compounds.

The use of anti-mycotoxin additive, liver protectors such as methionine and choline-feed shows some effect, especially in recovering the appetite of intoxicated animals. The use of natural additives or modified by the addition of compounds or biological enzymes requires further scientific study, but in field situations, some have been effective.

On a technical point of view, there are two criteria to take into account in order to define any product as an AMA: in vitro and in vivo evaluation results. However, to be released to market in Brazil, products must be registered in the Brazilian Department of Agriculture.

In vitro evaluation

The in vitro evaluation has to be run to determine the capability of the product to adsorb or inactivate mycotoxins present in a liquid medium and make them unavailable.

One product can show different adsorption rates in evaluations within different fluids. In fluids that mimic gastric/intestinal conditions product will face some pH conditions and the presence of some enzymes that are not present in hydro-alcoholic solutions.

With over 20 years of experience in mycotoxin analysis and more than 10 years evaluating AMA, LAMIC applies worldwide accepted methodologies to evaluate AMA in vitro.

Those methodologies use gastric and intestinal juices that accurately represent the conditions AMA will be submitted to in the animal.

There are two solutions prepared separately: gastric juice (pH 3.0) and intestinal juice (pH 6.0), both described in the Pharmacopeia National Formulary (1990). Those solutions are spiked with the mycotoxins intended to be adsorbed plus the AMA to be evaluated.

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The in vitro adsorption rate calculation is done based on the chromatographic response of the juice containing both mycotoxin and AMA, against the juice containing only the mycotoxin. The chromatographic response is obtained in the latest HPLC systems (HPLC/MS and/or HPLC/MS-MS).

LAMIC have evaluated more than 600 products from different origins (Germany, Argentina, Austria, Belgium, Brazil, Chile, Colombia, Cuba, Spain, Italy, Mexico, Pakistan, Peru, and USA).

For aflatoxin only, according to the results of those evaluations, more than 50% showed an in vitro adsorption rate higher than 90%, which means there is a great variety of products to be developed as AMA in vivo.

In vivo evaluation

Running an in vivo test requires more than just a laboratory and some equipment. Those tests must be run in specific environment and sanitary conditions. Thus, some experimental facilities are needed too.

The standard in vivo evaluation protocol has four treatments: One to control the experiment (negative control), another to control the product, the mycotoxin control (positive control), and the last one mixing the mycotoxin and the product (test group).

The evaluated parameters depend on the mycotoxin used, STI target organs and most important effects in the animal, but generally include performance parameters, biochemical

chemistry, and size and/or relative weight of organs. To set the product to an AMA group must show the test statistic significant difference compared with the positive control group. Fig. 2 presents an example of two products evaluated for zearalenone in pigs.

LAMIC evaluated 132 products and only 36% achieved the criteria established above.

Those data highlight the need to run suitable evaluations since nearly 64% of the products do not actually work as AMA.

Conclusion

The Brazilian experience with the management of mycotoxins has been associated with rapid growth of agribusiness. For the pace to be maintained and the business profitability to be preserved, technology had to be developed in parallel.

The main points were marked by the rapid assimilation of knowledge by the technical sector, by technology implementation, as the sampling and technology for analysis of mycotoxins compatible with efficiency that the market demands.

To solve the problem mycotoxicoses of both clinics and the only result in economic losses viewing difficult, as solutions to the AMAs had to be implemented, consequently generating the need for this class of products, ensuring their jobs safely and economically.

The strategies for storage control and good agricultural practices should be the next steps in this process, ensuring the economic excellence of the agribusiness and food safety. ■

Fig. 2. Results of in vivo evaluations of vulva volume (cm³) of gilts fed diets with and without AMA and zearalenone.

